Nota Lepi. **41**(1) 2018: 119–123 | DOI 10.3897/nl.41.23626

Large male bias in collection of *Micropterix facetella* Zeller, 1851 (Lepidoptera, Micropterigidae)

David Plotkin[1,2], Vazrick Nazari[3], Nicholas T. Homziak[1,2], Akito Y. Kawahara[1,2]

1 *Florida Museum of Natural History, University of Florida, Gainesville, FL, U.S.A.; dplotkin@ufl.edu*
2 *Department of Entomology and Nematology, University of Florida, Gainesville, FL, U.S.A.; nhomziak@ufl.edu, kawahara@flmnh.ufl.edu*
3 *Canadian National Collection of Insects, Arachnids and Nematodes; Ottawa Research and Development Centre, Agriculture and Agri-Food Canada, Ottawa, Ontario K1A 0C6, Canada; vazrick.nazari@agr.gc.ca*

http://zoobank.org/31DE8925-75CD-4BD7-98F9-1055BC7607C9

Received 16 January 2018; accepted 16 April 2018; published: 16 May 2018
Subject Editor: David C. Lees.

Micropterigidae are a small family of moths containing approximately 160 described species (van Nieukerken et al. 2011), the majority of which are in the type genus *Micropterix* (Kristensen 1998). Adult micropterigids possess mandibulate mouthparts, lacking the proboscis found in the more recently diverging glossatan lineages of Lepidoptera, and many micropterigid larvae are associated with non-angiosperm plants such as liverworts (Imada et al. 2011). For these reasons, extant micropterigids are frequently referred to as primitive moths. Recent phylogenetic analyses (e.g., Bazinet et al. 2016) have shown that the family Micropterigidae is indeed sister to all other Lepidoptera. Adults are usually diurnal and can be observed feeding on pollen or fern spores (Davis and Landry 2012; Kawahara et al. 2018). However, because of their small size, they are collected relatively infrequently.

In April 2017, while attending the 20th European Congress of Lepidopterology in southern Croatia, the authors collected butterflies and small moths near shrubs and oak trees (tentatively identified as *Quercus pubescens* Willd., 1796; Bašić 2013) on a south-facing rocky karst hillside northeast of the town of Podgora (Fig. 1; see below for specifics about location). Upon closer examination, many of the moths perched on the oaks were adult micropterigids (Fig. 2), though none of them were actually observed feeding at the inflorescences. Multiple trips were made to this site over the duration of the seven-day conference, with all collecting done in the afternoon, between the hours of 12:00 PM and 4:00 PM. There was also one attempt to collect at this site in the late morning, shortly after 10:00 AM, but no moths were observed. A total of 145 micropterigid specimens were collected by the authors. All specimens were later identified as *Micropterix facetella* (C. Zeller pers. comm.), which is one of seven micropterigid species known from Croatia (Karsholt 2013). The identification was further confirmed by DNA barcoding of a male specimen (CNCLEP000171989). Interestingly, approximately 80% of these specimens (117/145) were males.

Figure 1. Hillside near Podgora, Croatia, where *M. facetella* specimens were collected.

Figure 2. *Micropterix facetella*, habitus.

The underlying cause for this 4:1 sex ratio remains unclear, though there are multiple plausible explanations. Females may not fly as often as males, in order to preserve energy for creation of progeny. Alternatively, *Micropterix facetella* may be protandrous; this has never been officially demonstrated due to the difficulties of rearing Micropterigidae, but since there are many examples of males emerging before females in other species of Lepidoptera, it is not without precedent (Wiklund and Fagerström 1977). It is also possible that both sexes emerge simultaneously, but exhibit sexually dimorphic flight behavior. If additional collecting had been done in the morning or during twilight hours, a significantly different sex ratio may have been observed.

Observations of congeneric micropterigids indicate that the 4:1 sex ratio in *M. facetella* may be the result of a particular mating strategy. Kozlov and Zvereva (2006) studied adult activity of *Micropterix maschukella* Alphéraky, and observed that females visited host flowers primarily with the intent to feed, whereas males did so primarily with the intent to mate. These males preferred to visit flowers that were already occupied by other *M. maschukella* males, resulting in an observably biased sex ratio not dissimilar to the one encountered at the Croatian oaks. This sort of mating strategy, analogous to lekking, is rare in Lepidoptera, though it has been observed in some species from the families Nymphalidae (Srygley and Penz 1999), Pyralidae (Alem et al. 2011) and Hepialidae (Turner 2015). Similar male aggregations have been observed in other attempts to collect micropterigids (Zeller and Huemer 2015; D. Davis and D. Wagner pers. comm.). Gregarious behavior was also observed in the pollen-feeding *Micropterix calthella* Linnaeus (Erenler and Gillman 2010), though the sex ratio was not recorded. Despite these multiple corroborative studies, the variation in recorded flight time and behavior across all *Micropterix* is significant enough that, with regards to analyzing *M. facetella* behavior, it must all be treated as circumstantial (Zeller-Lukashort et al. 2007)

There are other known occurrences of gender bias in Lepidoptera. The bacterial endosymbiont *Wolbachia* has been found in many lepidopteran families and is known to kill male progeny during early development (Sasaki and Ishikawa 1999; Jiggins et al. 2000; Ahmed et al. 2015, 2016), creating an uneven sex distribution in favor of females. Gender bias towards females has also been observed in gypsy moth adults, due to some parasitoids preferring to parasitize male pupae (Fuester and Taylor 1996). These phenomena are less likely to be causing the 4:1 male-to-female ratio in *M. facetella*, but should not be entirely ruled out.

Although our observations of *M. facetella* are interesting on their own, it will require more than a single week of collecting in order to properly interpret them. Multiple avenues of additional research are needed in order to accurately infer the underlying cause of the observed male bias. Replicating the experimental designs of Kozlov and Zvereva (2006) with *M. facetella* would not only help determine whether the males are lekking, but would also help determine whether the females were actually feeding on oak flowers. This would also enable an assessment of whether the bias is strictly due to variation in diel activity (Kawahara et al. 2018). Rearing *M. facetella* would be necessary to test the protandry hypothesis, though there are relatively few instances of successful rearing of micropterigids from egg to adult (e.g., Carter and Dugdale 1982). Finally, the ethanol-preserved *M. facetella* specimens could be sequenced in order to test for the presence of *Wolbachia* DNA.

Material examined. 110 ♂, 28 ♀. CROATIA: Split-Dalmatia County, nr. Podgora; 43.256°N 17.086°E; 23–30.iv.2017; N.T. Homziak, A.Y. Kawahara, D.M. Plotkin (Molecular collection at the McGuire Center for Lepidoptera and Biodiversity, Florida Museum of Natural History, Gainesville, Florida); 7♂, same data, V. Nazari, A.Y. Kawahara (Canadian National Collection of Insects, Ottawa, Ontario, Canada).

Acknowledgements

We thank Christof Zeller (Museum Haus der Natur, Salzburg, Austria) for his assistance with identification of specimens, and Don Davis (USNM, Washington, D.C., U.S.A.) and David Wagner (University of Connecticut, Storrs, CT, U.S.A.) for providing additional information on micropterigids.

References

Ahmed MZ, Araujo-Jnr EV, Welch JJ, Kawahara AY (2015) *Wolbachia* in butterflies and moths: geographic structure in infection frequency. Frontiers in Zoology 12: 16. https://doi.org/10.1186/s12983-015-0107-z

Ahmed MZ, Breinholt JW, Kawahara AY (2016) Evidence for common horizontal transmission of *Wolbachia* among butterflies and moths. BMC Evolutionary Biology 16: 118. https://doi.org/10.1186/s12862-016-0660-x

Alem S, Koselj K, Siemers BM, Greenfield MD (2011) Bat predation and the evolution of leks in acoustic moths. Behavioral Ecology and Sociobiology 65: 2105–2116. https://doi.org/10.1007/s00265-011-1219-x

Bašić F (2013) The Soils of Croatia. World Soils Book Series, Springer, Dordrecht, 179 pp.

Bazinet AL, Mitter KT, David DR, Nieukerken EJ van, Cummings MP, Mitter C (2016) Phylotranscriptomics resolves ancient divergences in the Lepidoptera. Systematic Entomology 42: 305–316. https://doi.org/10.1111/syen.12217

Carter DJ, Dugdale JS (1982) Notes on collecting and rearing *Micropterix* (Lepidoptera: Micropterigidae) larvae in England. Entomologist's Gazette 33: 43–47.

Davis DR, Landry J-F (2012) A review of the North American genus *Epimartyria* (Lepidoptera, Micropterigidae) with a discussion of the larval plastron. ZooKeys 183: 37–83. https://doi.org/10.3897/zookeys.183.2556

Erenler HE, Gillman MP (2010). Synchronisation of adult activity of the archaic moth, *Micropterix calthella* L.(Lepidoptera, Micropterigidae), with anthesis of sedges (*Carex* spp., Cyperaceae) in an ancient wood. Arthropod-Plant Interactions 4: 117–128. https://doi.org/10.1007/s11829-010-9090-7

Fuester RW, Taylor PB (1996) Differential mortality in male and female gypsy moth (Lepidoptera: Lymantriidae) pupae by invertebrate natural enemies and other factors. Environmental Entomology 25: 536–547. https://doi.org/10.1093/ee/25.2.536

Imada Y, Kawakita A, Kato M (2011) Allopatric distribution and diversification without niche shift in a bryophyte-feeding basal moth lineage (Lepidoptera: Micropterigidae). Proceedings of the Royal Society of London B: Biological Sciences: rspb20110134. https://doi.org/10.1098/rspb.2011.0134

Jiggins FM, Hurst GD, Dolman CE, Majerus ME (2000) High prevalence male-killing *Wolbachia* in the butterfly *Acraea encedana*. Journal of Evolutionary Biology 13: 495–501. https://doi.org/10.1046/j.1420-9101.2000.00180.x

Karsholt O (2013) Fauna Europaea: Lepidoptera, Micropterigidae. Fauna Europaea version 2017.06 https://fauna-eu.org [accessed on 05.12.2017]

Kawahara AY, Plotkin D, Hamilton CA, Gough H, St Laurent R, Owens HL, Homziak NT, Barber JR (2018) Diel behavior in moths and butterflies: a synthesis of data illuminates the evolution of temporal activity. Organisms Diversity & Evolution 18: 13–27. https://doi.org/10.1007/s13127-017-0350-6

Kristensen NP (1998) The non-glossatan moths. In: Kristensen NP (Ed.) Lepidoptera, Moths and Butterflies, 1 Evolution, systematics and biogeography. Handbuch der Zoologie/ Handbook of Zoology 35. De Gruyter, Berlin, New York, 41–49. https://doi.org/10.1515/9783110804744.41

Kozlov MV, Zvereva EL (2006) Aggregation of *Micropterix maschukella* moths on inflorescences of common elder: mating at foraging sites (Lepidoptera Micropterigidae). Ethology Ecology & Evolution 18: 147–158. https://doi.org/10.1080/08927014.2006.9522719

Nieukerken EJ van, Kaila L, Kitching IJ, Kristensen NP, Lees DC, Minet J, Mitter C, Mutanen M, Regier JC, Simonsen TJ, Wahlberg N, Yen S-H, Zahiri R, Adamski D, Baixeras J, Bartsch D, Bengtsson BÅ, Brown JW, Bucheli SR, Davis DR, de Prins J, de Prins W, Epstein ME, Gentili-Poole P, Gielis C, Hättenschwiler P, Hausmann A, Holloway JD, Kallies A, Karsholt O, Kawahara AY, Koster S, Kozlov M, Lafontaine JD, Lamas G, Landry JF, Lee S, Nuss M, Park K-T, Penz C, Rota J, Schintlmeister A, Schmidt BC, Sohn J-C, Solis MA, Tarmann GM, Warren AD, Weller S, Yakovlev RV, Zolotuhin VV, Zwick A (2011) Order Lepidoptera Linnaeus, 1758. In : Zhang Z-Q (Ed.) Animal biodiversity: an outline of higher-level classification and survey of taxonomic richness. Zootaxa 3148: 212–221.

Sasaki T, Ishikawa H (1999) *Wolbachia* infections and cytoplasmic incompatibility in the almond moth and the Mediterranean flour moth. Zoological Science 16: 739–44. https://doi.org/10.2108/zsj.16.739

Srygley RB, Carla MP (1999) Lekking in neotropical owl butterflies, *Caligo illioneus* and *C. oileus* (Lepidoptera: Brassolinae). Journal of Insect Behavior 12.1: 81–103. https://doi.org/10.1023/A:1020981215501

Turner JR (2015) The flexible lek: *Phymatopus hecta* the gold swift demonstrates the evolution of leking and male swarming via a hotspot (Lepidoptera: Hepialidae). Biological journal of the Linnean Society 114: 184–201. https://doi.org/10.1111/bij.12411

Wiklund C, Fagerström T (1977) Why do males emerge before females? Oecologia 31: 153–158. https://doi.org/10.1007/BF00346917

Zeller HC, Huemer P (2015) A new species of *Micropterix* Hübner, 1825 from the Orobian Alps (Italy)(Lepidoptera, Micropterigidae). Nota Lepidopterologica 38: 133–146. https://doi.org/10.3897/nl.38.5058

Zeller-Lukashort HC, Kurz ME, Lees DC, Kurz MA (2007) A review of *Micropterix* Hübner, 1825 from northern and central Europe (Micropterigidae). Nota Lepidopterologica 30: 235–298.